**Supplemental Figure S1.** Distributions of loop fluctuations similar to Figure 2 in the paper, for the additional proteins (A) 1gjn, (B) 1amk, and (C) 101w. See Figure 2 caption for further details.



**Supplemental Figure S2.** Mobility plots similar to Figure 3 for (A) 1GJN, (B) 1AMK, and (C) 1O1W. For colored images, the helices are colored red, strands yellow, and loops gray.



**Supplemental Figure S3.** Plots of PCA percent of variance in the first PC, similar to Figure 5, but for the additional proteins (A) 1GJN, (B) 1AMK, and (C) 1O1W. See Figure 5 caption for further details.



**Supplemental Figure S4.** Dot products between PCs computed from free peptides of length (A) 4, (B) 7, and (C) 9 and PCs from the same loop in 1flh. In panel A, it may appear that the motion space is similar, but that the modes have simply been shuffled in one ensemble relative to the other. This, however, is not the case since each PC has an important weight factor; percent of captured variance. Thus, the direction of PC1 of the 4 amino acid loop in 1flh has an overlap of 0.77 with the free peptide's 9th PC, indicating a high directional agreement, but the shift from PC1 to PC9 indicates that the direction of motion that is most dominant in the full structure is trivially used in the free peptide. Thus the overall motion that combines all of the modes of motion will be substantially different.



Length	aa	ALA
4	0.29	0.39
7	1.08	0.98
9	0.96	1.06
15	1.07	1.11

**Supplemental Table S1.** Similarity of loop models between free peptides and end-restrained using WRMSIP.

WRMSIP between the two null models; the free peptide and a simulation where the alpha carbons of the N- and C-terminal residues were harmonically constrained with a force of 5 kcal/mol. WRMSIPs are calculated between each null model simulation. We find that the motions of the short loop are significantly altered by end constraints, compared to the unconstrained simulation, while longer loops show similar motions. We wanted WRMSIP to be commutative, but this alters the range of WRMSIP compared to RMSIP which is [0, 1]. Values above 1 indicate that the trajectories are not only sampling similar motions, but the extent of sampling is shifted towards the PCs that capture a higher percent of variance. 'aa': the simulation was performed for the 1FLH sequence listed below; 'ALA': a poly-alanine chain of the same length was simulated.

## PCA of Individual Loops from the Full Trajectory

1lh.pdb											
Residues	Length	#exposed	PC1	PC2	PC3	PC4	PC5	first5			
10:12	3	2	65.00	18.99	11.21	3.16	1.10	99.46			
21:24	4	0	49.09	29.26	18.67	1.45	0.94	99.41			
33:37	5	3	68.36	15.61	7.82	2.81	2.02	96.62			
43:47	5	2	46.92	40.87	6.90	3.26	1.45	99.41			
52:55	4	3	61.82	25.65	6.83	2.98	2.14	99.41			
61:64	4	1	52.49	36.70	6.60	2.66	0.90	99.37			
66:69	4	4	47.76	41.06	6.37	2.35	1.51	99.05			
75:78	4	0	54.30	35.55	8.32	1.15	0.36	99.68			
107:110	4	2	50.35	37.57	6.54	3.33	1.43	99.21			
115:118	4	4	57.61	22.71	16.60	2.06	0.59	99.57			
123:125	3	3	56.63	17.91	14.07	7.57	2.07	98.25			
133:135	3	3	39.92	29.42	18.81	6.10	3.88	98.12			
143:149	7	4	40.02	22.12	17.03	12.00	3.79	94.96			
155:163	9	9	54.43	28.19	7.00	5.03	2.99	97.64			
168:171	4	4	49.40	28.67	11.98	7.33	1.41	98.78			
176:179	4	1	48.20	24.63	12.77	9.66	2.29	97.54			
184:186	3	3	51.16	30.46	10.53	5.19	1.78	99.13			
206:209	4	1	68.62	20.80	5.51	3.66	0.66	99.27			
215:220	6	6	77.92	9.17	6.35	2.92	1.16	97.51			
235:237	3	0	57.46	24.13	13.29	3.29	1.01	99.17			
240:244	5	4	55.65	32.53	5.83	4.49	0.70	99.21			
255:257	3	0	61.94	20.14	12.05	3.88	0.98	98.98			
278:281	4	2	71.14	15.97	6.78	3.44	1.29	98.62			
289:291	3	3	75.86	12.17	8.64	1.62	0.89	99.20			
315:318	4	2	61.58	20.46	9.04	6.61	1.50	99.20			
CC(n,cPC1) =	-0.09	% n is	the loc	op lengt	th and 1	ne is the	e #expo	sed residues			
CC(ne,cPC1)	= 0.04	% c1-3	3 is the	total a	CC(ne,cPC1) = 0.04 % c1-3 is the total across the first 3 PCs						

CC(n,c1-3) = 0.24

CC(ne,c1-3) = 0.19

% c1-3 is the total across the first 3 PCs % CC stands for Pearson's correlation coefficient

## 1gjn.pdb

Residues	Leng	gth #exposed	PC1	PC2	PC3	PC4	PC5	first5
1:3	3	3	63.90	16.86	10.88	5.76	1.76	99.17
41:51	11	9	38.66	15.15	13.19	12.04	9.09	88.14
77:82	6	4	44.31	30.92	13.20	3.83	2.97	95.24
96:100	5	4	49.60	27.69	11.72	4.75	4.19	97.95
119:124	6	3	42.44	27.51	10.98	9.01	4.93	94.87
150:153	4	3	39.69	25.79	20.58	10.55	2.81	99.43
CC(n,cPC1	) = -0.62	2						
CC(ne,cPC	(1) = -0.4	42						
CC(n,c1-3)	= 0.53							
CC(ne,c1-3	) = 0.50							

## 1amk.pdb

Residues	Length	n#exposed	PC1	PC2	PC3	PC4	PC5	first5
1:6	6	5	59.80	27.88	4.46	3.55	1.60	97.30
12:17	6	2	55.79	17.86	10.03	8.33	3.01	95.02
30:37	8	3	64.12	15.44	7.34	4.92	2.85	94.67
55:59	5	3	56.97	25.45	6.91	5.26	3.35	97.94
65:67	3	1	76.59	8.16	7.55	2.62	2.23	97.14
69:78	10	3	54.35	21.98	8.09	5.18	3.27	92.85
86:89	4	3	54.28	27.27	9.88	3.79	2.14	97.37
102:105	4	4	70.61	18.24	5.54	2.50	1.67	98.56
119:121	3	2	46.37	30.31	14.76	5.27	1.79	98.50
128:130	3	2	50.46	32.72	11.60	2.95	1.49	99.22
136:138	3	2	57.30	29.43	7.69	3.52	1.35	99.30
152:155	4	3	57.55	15.55	10.99	7.08	4.47	95.64
172:179	8	7	86.97	7.43	2.21	1.36	0.86	98.84
212:218	7	4	43.47	25.90	14.35	9.03	3.87	96.61
224:229	6	3	52.77	24.22	8.06	7.03	3.32	95.41
239:241	3	0	49.10	27.07	14.25	4.87	3.73	99.02
248:250	3	3	61.00	20.30	10.71	5.91	1.47	99.40
CC(n,cPC1) =	0.15							

CC(ne,cPC1) = 0.47CC(n,c1-3) = 0.18

CC(ne,c1-3) = -0.21

1o1w.pdb								
Residues	Length	#exposed	PC1	PC2	PC3	PC4	PC5	first5
1:15	15	11	73.89	19.80	4.03	1.81	0.48	100.00
38:40	3	3	88.89	8.75	2.34	0.02	0.00	100.00
45:51	7	3	62.75	32.18	4.60	0.33	0.14	100.00
66:69	4	1	81.62	15.61	2.19	0.53	0.06	100.00
83:94	12	3	88.89	7.33	2.27	0.98	0.53	100.00
105:107	3	1	56.74	39.45	3.19	0.58	0.04	100.00
113:138	26	16	48.54	32.55	11.16	4.47	3.28	100.00
CC(n,cPC1) =	-0.46							
CC(ne,cPC1) = -0.49								
CC(n,c1-3) = 0.96								
CC(ne,c1-3) = 0.93								

## 1flh Fragments Used In Individual Loop Studies

These are the same loops that are simulated independently and presented in Table 2. All three smaller loops are surface exposed, while the 15aa fragment is a buried strand that connects two surface exposed loops.

4aa sequence:	GTPA	Residues: 21-24
7aa sequence:	QGLVSQD	Residues: 143-149
9aa sequence:	LSADDQSGS	Residues: 155-163
15aa sequence:	TGSSNLWVPSVYCSS	Residues: 33-47